ANEMIA OF COWS IN THE ZONE OF RADIOACTIVE CONTAMINATION OF THE WESTERN REGION OF UKRAINE

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INTRODUCTION

One of the grave consequences of the accident at the Chornobyl NPP was the radioactive contamination of a large territory of Ukraine, including agricultural land. According to official data, more than 50 million curies of various radionuclides were released into the environment. However, individual scientists estimate this figure to be much higher – about 1 billion curies. The scale of radioactive contamination is huge – 46 percent of the territory of Ukraine has contamination higher than 37 kBq/m² (1 Ki/km²). Most regions of Ukraine are polluted. The districts of Kyiv, Zhytomyr, Chernihiv, Rivne, and Volyn regions were the most affected^{1,2,3}.

1. The radiation situation in certain districts of the Rivne region

The main ecological changes took place in the territories with well-developed animal husbandry, and the consequences of the disaster were aggravated by the fact that the active phase of the accident at the Chornobyl NPP was more than 10 days. During this time, due to the change in the wind direction, several radioactive traces were formed, covering large areas, particularly the Rivne region, which was exposed to radiation and belongs to the western biogeochemical zone of Ukraine (Tables 1–3)⁴.

According to the Decree of the Cabinet of Ministers of Ukraine, 5 settlements of the Dubrovytskyi district, including 2 collective farms, are assigned to the second zone of radioactive contamination from the

² 20 років Чорнобильської катастрофи. *Погляд у майбутнє: Національна доповідь* України. Київ : Атіка, 2006. 224 с.

¹ Малиновський А. С. Еколого-економічні та соціальні аспекти Чорнобильської катастрофи (на прикладі Житомирської області). Київ, 2001. С. 9–14; 234–236.

³ Левченко В.І., Слівінська Л.Г. Анемія у корів зони техногенного забруднення. *Наук. вісник. вет. медицини.* Біла Церква, 2010. Вип. 5(78). С. 102–111.

⁴ Дмитрієв В., Романюк М., Ситарчук В. Мікроелементози та їх профілактика на Рівненщині. *Вет. медицина України*. 2003. № 10. С. 19–20.

territory of the Rivne region; to the third -269 settlements of five districts (99 collective farms); to the fourth -64 settlements of three districts (24 collective farms). A total of 338 settlements are located on the region's polluted territory. The results of the density of soil pollution in the Rivne region are shown in the table. 4.

It is worth mentioning that sod-podzolic and peat-swamp soil with a high level of surface groundwater and high acidity are the most common in the farms of the Rivne region. The soils are poor in calcium, manganese, and one and a half oxides of aluminum and ferrum.

As a result of the decay of short-lived radionuclides, radioactive contamination of the territory is determined by two long-lived ones: cesium-137 and strontium-90, which have a half-life of almost 30 years⁵. They fell primarily in the condensation form composition, which is the most accessible for plant absorption. But over time, cesium-137 is gradually fixed on parts of the solid phase of the soil, and now it's "aging" is taking place – a decrease in its availability for plant root nutrition. That is why in the 15 years since the accident at the Chornobyl NPP, the specific radioactivity of cesium-137 has significantly decreased in plants (by 2–3 times without the use of countermeasures), while strontium-90 is in the soil and therefore intensively enters plants.

Table 1
The area of ¹³⁷Cs contamination of agricultural land in the regions of the western region of Ukraine (thousand ha)

Regions		Distribution by pollution density, kBq/m² (Ki/km²)					
	3,7-37 (0,1-1)	37–185 (1–5)	37–185 (5–15) peat	185-555 (5-15)	185-555 (5-15)peat	More than 555(15)	
Volyn	228,4	15,4	10,2	0,2	_	_	
Ivano- Frankivsk	71,3	19,1	_	1,0	-	_	
Rivne	172,1	145,7	48,8	11,5	4,1	_	
Ternopil	219,3	12,5	_	_	_	_	
Chernivtsi	74,5	22,7	_	0,3	_	_	

Note. Indicators of the national report of Ukraine – "20 years of the Chernobyl disaster. A look into the future"⁶.

 $^{^5}$ Дмитрієв В., Романюк М., Ситарчук В. Мікроелементози та їх профілактика на Рівненщині. Вет. медицина України. 2003. № 10. С. 19-20.

⁶ Ведення сільського господарства в умовах радіоактивного забруднення території України внасділок аварії на Чорнобильській АЕС на період 1999–2002 рр. (методичні рекомендації) / Пристер Б. С., Кашпаров В. О., Надточій П. П. та ін. / Київ, 1998. 103 с.

	Distribution by pollution density, kBq/m ²					
Pollution density 137Cs, Ki/km² (kBq/m²)	Arable land, thousand hectares	In percentages	Hayfields and pastures, thousand ha	In percentages		
0,1-1 (3,7-37)	75,7	47,88	56,6	43,3		
1-5 (37-185)	47,8	30,23	37,6	28,8		
2-5 (74-185)	30,6	19,35	28,9	22,1		
5-10 (185-370)	3,9	2,48	7,1	5,4		
10-50 (370-1850)	0,1	0,06	0,5	0,4		
In total	158,1	100	130,7	100		

Examinations conducted by the radiological department of the Rivne Regional State Laboratory of Veterinary Medicine showed that radioactive cesium is the main component of pollution in the region. 288.8 thousand hectares of land were surveyed, including 158.1 thousand hectares of arable land and 130.7 thousand hectares of hayfields and pastures in six districts of the region.

The research was conducted on dry cows, which were born and constantly kept on farms in the Dubrovytskyi district of the Rivne region. The farm is located in the Northern part of the region, the territory of which, according to the Decree of the Cabinet of Ministers of Ukraine No. 106 of July 23, 1991, belongs to the third zone of radioactive contamination (Table 3). The indicators were compared with those of the cows of the Radyvyliv district of the Rivne region, located in the Southern part of the Rivne region.

Animal husbandry in the contaminated zone specializes in producing meat and milk. Cattle of the black and spotted breed are bred. Keeping cows is a stable pasture. Rough, juicy, and partially concentrated fodder is produced on the farm.

Radionuclides enter the body of animals during external irradiation – through the skin, mucous membranes, and lungs, inhalation of polluted air, and through the digestive tract with feed and water. Contamination of fodder that was fed to cows on the farm was: hay (47.2–320 Bq/kg), straw (32.1–455), silage (15–40), compound fodder (20–37), fodder beet and wood chips – 15–25 and 12–30 Bq/kg, respectively.

Density of contamination of agricultural land ¹³⁷Cs Dubrovytskyi district

Kinds of agricultural land	Pollution density, Ki/km² (kBq/m²)	Area, ha
	0,1-1 (3,7-37)	662,50
Arable land, ha	1- (37-185)	254,60
	5-15 (185-555)	_
Tota	917,10	
	0,1-1 (3,7-37)	1341,90
Hayfields and pastures	1-5 (37-185)	937,50
	5-15 (185-555)	_
Total hay	2279,40	

The value of the gamma background on the territory of the experimental farm ranged from 12 to 18 μ R/h.

The main share in livestock product contamination is ¹³⁷ Cs and ⁹⁰ Sr. The arrival of these elements in the body of animals occurs mainly due to their transition from the soil to plants and further – to the products of plant and animal husbandry⁷. Radionuclides migrate from the earth's surface with atmospheric precipitation to the lower soil layers, causing fodder accumulation. The main reasons for this situation are small amounts of improved land for grazing and forage harvesting.

Thus, animals born and kept on the territory of this farm and settlements were exposed to both external and internal radiation.

We compared the contamination of fodder on the farm of the contaminated zone, where the work was performed, and the conditionally clean zone. Based on the determination of the contamination of the ration fed to animals during the winter-stall period, we calculated the total daily intake of radionuclides cesium-137 and strontium-90 (tables 4, 5). The total diet activity for cows in radiostrontium (90 Sr) per day was 5.596 kBq 137 , Cs – 19.61 kBq.

The milk contamination with radionuclides in the experimental farm ranged from 15 to 60 Bq/kg. However, in the private sector, the maximum contamination of individual samples was 173–312 Bq/kg (the norm is no more than 100), which is explained by livestock grazing and harvesting of hay on forest lands, where, as a rule, the density of soil contamination is higher.

⁷ Слівінська Л. Г., Левченко В. І. Клінічний статус і показники гемопоезу лактуючих корів, вирощених на території, забрудненій радіонуклідами. *Сільський господар.* 2007. № 11–12. С. 33–36.

The name of the feed	Mass of feed consumed per day, kg	Specific radioactivity, kBq/kg	In total, cesium- ¹³⁷ was received with feeds, kBq
Cereal hay	3	2,53	7,59
Wheat straw	4	0,49	1,96
Corn silage	10	0,32	3,2
Soilage	6	0,57	3,42
Fodder beet	5	0,41	2,05
Corn grits	2	0,48	0,96
Sunflower meal	1,0	0,43	0,43
Total for a day			19,61

The content of radionuclides in the parenchymal organs of slaughtered animals ranged from 37 to 320 Bq/kg (generally up to 200). At the same time, animals from the individual sector had higher indicators.

On the farm, in addition to constant exposure to small doses of radioactive radiation, cows do not receive microelements and nutrients in the winter. As a result of long-term hypodynamia, with energy, vitamin, and mineral deficiency, the chronic effect on animals of low-intensity radioactive radiation, which reaches 5 Ki/km², leads to metabolic disorders.

Table 5
Contamination of cows' diets with radiostrontium-90 in the farm
of Dubrovytskyi district

The name of the feed	Mass of feed consumed per day, kg	Specific radioactivity, kBq/kg	In total, Sr ⁹⁰ arrived with feeds, kBk
Cereal hay	3	0,257	0,771
Wheat straw	4	0,189	0,756
Corn silage	10	0,062	0,62
Soilage	6	0,485	2,91
Fodder beet	5	0,041	0,205
Corn grits	2	0,113	0,226
Sunflower meal	1,0	0,108	0,108
Total for a day			5,596

The region's soils are characterized by a low content of mobile forms of iodine, copper, zinc, and cobalt^{8, 9}. Our research shows that the soils of the district are low in copper and cobalt: their content was 1.96 and 1.40 mg/kg, respectively.

When analyzing the diet (Table 6), we found that the content of dry matter and fiber exceeded the needs of beef cows by 23.9 and 64.2%, respectively. The digestible protein content in the cows' ration was 1035 g, if necessary, 1090 g. The sugar: digestible protein ratio was 0.82:1 (0.9:1 according to norms), but their provision in the diet was only 86.4 and 95.04 % until needed, respectively.

Table 6
The content of nutrients and biologically active substances in the diet of dry cows in the farm of the contaminated zone

Indexes	Units of measu- rement	In diet	Necessary	± Before neces- sary	In % to the necessary
Dry matter	kg	14,4	11,6	2,78	124,0
Exchange energy	MJ	123,0	116	7,0	106,0
Fodder units		9,85	9,9	-0,04	99,6
Raw protein	g	1680	1675	5	100,3
Digestible protein	g	1035,9	1090	-54,10	95,0
Cellulose	g	4385,0	2670	1715,0	164,2
Starch		1040	1175	-135	88,5
Sugar	g	847,0	980	-133,0	86,4
Calcium	g	94,5	95	-0,50	99,5
Phosphorus	g	57,3	55	2,3	104,2
Ferum	mg	4197,0	695	3502,0	604,0
Kopper	mg	82,3	100	-17,7	82,3
Zinc	mg	382,0	495	-113,0	77,2
Cobalt	mg	3,44	6,9	-3,46	49,9
Manganese	mg	1099,0	495	604,0	222,0
Iodine	mg	4,09	6,9	-2,81	59,3
Carotene	mg	390,0	495	-105,0	78,8

Analyzing the diet of the cows of the Dubrovytskyi district of the Rivne region in the winter period, a deficiency of trace elements, in particular copper and cobalt, was established, the availability of which was 82.3 and

⁹ Дмитрієв В., Романюк М., Ситарчук В. Мікроелементози та їх профілактика на Рівненщині. Вет. медицина України. 2003. № 10. С. 19–20.

 $^{^{8}}$ Мікроелементози сільськогосподарських тварин / М. О. Судаков, В. І. Береза, І. Г. Погурський [та ін.] ; за ред. М. О. Судакова. 2-е вид. Київ : Урожай, 1991. 144 с.

49.9%, and their concentration per 1 kg of dry matter was 5.7 and 0.24 mg (according to the norms of 8–10 and 0.55–0.80 mg, respectively (Table 6).

Thus, the animals that were born and kept on the farm were exposed to constant external and internal radiation exposure, which was supplemented by the negative impact of a deficiency of Co, Cu, zinc, iodine, digestible protein, starch, and sugar with a significant excess of ferrum (291 mg/kg of dry matter).

It is essential to mention that in the zone of radioactive contamination conditions, many studies were conducted to study the influence of trace elements Co, I, Cu, Mn, Zn, and others on productivity, resistance, mineral exchange, and contamination of livestock products with radionuclides ^{10, 11, 12, 13}. The issue of studying the influence of individual trace elements, particularly Co, Cu, and Fe, on erythrocyte poiesis in cows and their supplements' effectiveness in the polluted zone's specific conditions has not been studied enough.

2. Clinical status and indicators of hematopoiesis in cows in the zone of radiation contamination

During the clinical examination of 56 cows in the zone of radiation contamination, it was established that most animals had average fatness and dull hair cover. In 38 cows (63.8%), dryness and reduced skin elasticity were observed. In 20 (35.7%), the conjunctiva was pale pink, and the nose and mouth mucous membranes were anemic. During the cardiovascular system examination, the first heart sound was increased in 18 cows (32.1%), and tachycardia was recorded in 24 animals (42.9%).

For the analysis of erythrocytopoiesis indicators, blood was taken from 40 cows aged 3–6 years. The control was 20 cows aged 3–6 years from a conditionally clean zone.

Hemoglobin content in the blood of cows of the experimental and control groups differed significantly. Thus, in the cows of the experimental group, its level was, on average, 86.4±1.72 g/l and was probably (p<0.001) lower than

¹¹ Козенко О. В. Фізіологічний статус великої рогатої худоби за умов впливу абіотичних чинників середовища : автореф. дис. на здобуття наукового ступеня д-ра с.-г. наук : спец. 03.00.13 "Фізіологія людини і тварини" 16.00.06 "Гігієна тварин та ветеринарна санітарія". Львів, 2004. 41 с.

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¹⁰ Жила М. І. Морфологічна характеристика органів імунної системи молодняка великої рогатої худоби, вирощеної на радіоактивно забрудненій місцевості : автореф. дис. на здобуття наукового ступеня канд. вет. наук: спец. 16.00.02 "Патологія, онкологія і морфологія тварин". Київ, 2002. 19 с.

¹² Лігоміна І. П., Влізло В. В., Нікітенко А. М., Козак М. В. Вплив імуномодуляторів на неспецифічну резистентність телят в умовах тривалої дії радіації. *Наук. вісник Львів. держ. акад. вет. медицини імені С. З. Ґжицького.* Львів. 1999. Т. 9, № 3(34). Ч. 1. С. 143–145.

¹³ Слівінська Л. Г., Щербатий А. Р. Діагностика мікроелементозів кобил у західній біогеохімічній зоні України. *Вет. медицина України*. 2013. № 4(206). С. 25–28.

in the cows of the control group (115.3±2.11 g/l; Table 7). Oligochromemia was recorded in 26 (65%) cows studied in the zone of radiation contamination, while the hemoglobin content of cows in the conditionally clean zone was within the physiological range ^{14, 15, 16, 17}.

Table 7
Indicators of hematopoiesis in cows raised on the territory,
contaminated with radionuclides (M±m)

Indicator	Biometric indicator	Radyvylivskyi district (control) n=20	Dubrovytskyi district (study) n=40	p<
Hemoglobin, g/l	Lim M±m	95,0-127,0 115,3±2,11	69,0–107,0 86,4±1,72	0,001
Erythrocytes, T/l	Lim M±m	5,2-7,4 6,5±0,14	3,3-5,2 4,2±0,10	0,001
MCH, pg	Lim M±m	16,2–19,8 17,9±0,23	17,9–24,4 20,6±0,25	0,001
HCT, in percent	Lim M±m	28,0-35,0 31,7±0,48	22,0-32,0 25,7±0,35	0,001
MCV, μm3	Lim M±m	41,7–61,1 49,4±1,22	46,2–73,8 61,7±0,95	0,001

Notes: p< – compared cows of the zone of radiation contamination (experiment) and conditionally clean zone (control).

The number of erythrocytes in the cows of the experimental group ranged from 3.3 to 5.2 T/l, averaged 4.2±0.10 T/l, and was 35.4% less (p<0.001) compared to the animal control group. Oligocythemia was established in 36 cows (90%). In 26 cows (65%), oligocythemia was combined with

 14 Слівінська Л. Г. Левченко В. І. Клінічний статус і показники гемопоезу лактуючих корів, вирощених на території, забрудненій радіонуклідами. *Сільський господар.* 2007. № 11–12. С. 33–36.

¹⁶ Слівінська Л.Г. Еритроцитопоез та обмін заліза у тільних корів. *Вісник Білоцерків. держ. аграр. ун-ту.* 2006. № 40. С. 182–188.

Riga, Latvia : «Baltija Publishing». 2023, 136–164.

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¹⁵ Slivinska, L., Shcherbatyy, A., Gutyj, B., Lychuk, M., Fedorovych, V., Maksymovych, I., Rusyn, V., Chernushkin, B. Parameters of erythrocytopoiesis, acid resistance and population composition of erythrocytes of cows with chronic hematuria. *Ukrainian journal of Ecology*. 2018. Vol. 8. Iss. 1. P. 379–385. Doi: 10.15421/2018_225.

¹⁷ Slivinska L. G., Fedorovych V. L., Shcherbatyi A. R. Effectiveness of inorganic and chelate compounds of microelements for osteodystrophy of cows. *Achievements and research prospects in animal husbandry and veterinary medicine*: Scientific monograph.

oligochromemia (Table 8). In 10 (25%), it occurred independently. Thus, anemia was diagnosed in 36 cows (90% of the examined).

The content of hemoglobin in erythrocytes (RBC) in the blood of cows in the polluted zone ranged from 17.9 to 24.4 pg and averaged 20.6 ± 0.25 pg and was probably 15% higher (p<0.001) than the control (17.9±0.23 pg). The analysis of the blood tests of cows with anemia showed that anemia is hyperchromic in 22 (6.1%) cows.

An important indicator characterizing the state of erythrocytopoiesis is the value of the hematocrit. Under the influence of low doses of radiation, this indicator in cows was, on average, $25.7\pm0.35\%$, while in the control, it was $31.7\pm0.48\%$. In cows of the conditionally clean zone, the hematocrit value was probably (p<0.001) higher by 6%. The decrease in the hematocrit value occurred as a result of oligocythemia since the average volume of erythrocytes (MCV) in cows of the radiation-contaminated zone was significantly (p<0.001) greater by 25% than in cows of the conditionally clean zone, and on average was $61.7\pm0.95~\mu m^3$. In 24 cows, the MCV was greater than $60~\mu m^3$. The volume of erythrocytes in them ranged from $61.0~to~73.8~\mu m^3$, while in cows of the conditionally clean zone $-~41.7.5-61.1~\mu m^3$. Characteristics of anemias are given in Table 8.

Analyzing the blood test results we recorded, the negative effect of low ionizing radiation on the body can be traced in cows in the radiation pollution zone, as a decrease in erythrocytes, hemoglobin content, and hematocrit value is observed.

And yet, an essential reason for the development of macrocytic hyperchromic anemia in cows is not only the effect of low doses of ionizing radiation on the hematopoietic system but also cobalt deficiency, characteristic of hypoplastic anemia.

Table 8
Characteristics of anemia in cows raised
in the territory contaminated with radionuclides

Changes in indicators		Including			
of erythrocytopoiesis	In total	hyper- chromic	normo- chromic	macro- cytic	normo- cytic
Anemia	$\frac{36}{90}$	-	-	_	-
Oligocythemia	$\frac{36}{90}$	<u>22</u> 61,1	$\frac{14}{38,9}$	$\frac{24}{66,7}$	$\frac{12}{33,3}$
Oligochromemia	$\frac{26}{65}$	$\frac{12}{33,3}$	$\frac{14}{38,9}$	17 47,2	$\frac{9}{25}$
Oligocythemia	$\frac{26}{65}$	12 33,3	14 38,9	$\frac{17}{47,2}$	$\frac{9}{25}$

Note: in the numerator – total cows, in the denominator – in percent.

3. Acid resistance and age composition of erythrocytes of cows in the zone of radiation contamination

Studying the age of erythrocytes is worthy of attention from the point of view of the study of bone marrow functions. As the cells age, their lipoproteins become depleted, the sulfhydryl and peroxidase activity of the protoplasm decreases, and the content of histidine and lipids partially changes^{18, 19, 20, 21}. In such cells, lipid peroxidation intensity increases, leading to destructive processes in plasma membranes and disrupting the transport of cations and amino acids. In cows located in the zone of radioactive contamination, the proportion of "young" erythrocytes ranged from 42.7 to 53.6 and averaged 48.1±1.15%. In contrast, in cows of the control group, it was 44.1±0 .35% (p<0.01), ranging from 42.6 to 45.8% (Table 9). It is evident that with long-term radiation exposure, the bone marrow is irritated, which causes the influx of "young" cells into the blood, which is aimed at eliminating oxygen starvation.

Table 9
Indicators of the population composition of erythrocytes in the blood of cows raised in the territory contaminated with radionuclides

A group	"0	"Old" "Mature" "Young"		"Mature"		ıng"
of cows	in percent	T/l	in percent	T/l	in percent	T/I
Control (n= 20)						
Lim	10,7-14,5	0,73-1,02	41,7-45,4	1,65-2,16	42,6-45,8	1,36-1,96
M±m	12,5±0,33	$0,8\pm0,03$	$43,4\pm0,4$	$2,8\pm0,06$	$44,1\pm0,35$	$2,9\pm0,06$
Дослідна (n= 40)						
Lim	10,7–17,4	0,47-0,74	32,6-42,4	1,40-1,89	42,7-53,6	1,63-2,06
M±m	14,2±0,51	$0,58\pm0,02$	$37,7\pm0,76$	$1,56\pm0,04$	$48,1\pm1,15$	$2,0\pm0,13$
p<	0,05	0,001	0,001	0,001	0,01	0,001

Note: p< – compared to the control group.

²⁰ Слівінська Л.Г. Структурно-функціональні властивості еритроцитів за анемії різної етіології. *Вісник Білоцерків. нац. ун-ту.* 2009. № 62. С. 81–87.

¹⁸ Slivinska, L., Shcherbatyy, A., Gutyj, B., Lychuk, M., Fedorovych, V., Maksymovych, I., Rusyn, V., Chernushkin, B. Parameters of erythrocytopoiesis, acid resistance and population composition of erythrocytes of cows with chronic hematuria. *Ukrainian journal of Ecology*. 2018. Vol. 8. Iss. 1. P. 379–385. Doi: 10.15421/2018_225.

¹⁹ Slivinska, L. G., Shcherbatyy, A. R., Lukashchuk, B. O., Zinko, H. O., Gutyj, B. V., Lychuk, M. G., Chernushkin, B. O., Leno, M. I., Prystupa, O. I., Leskiv, K. Y., Slepokura, O. I., Sobolev, O. I., Shkromada, O. I., Kysterna, O. S., Usiienko, O. V. Correction of indicators of erythrocytopoesis and microelement blood levels in cows under conditions of technogenic pollution. *Ukrainian journal of Ecology.* 2019. Vol. 9. Iss. 2. P. 127–135.

²¹ Golovakha, V. I., Piddubnyak, O. V., Sliusarenko, S. V., Slivinska, L. G., Maksymovych, I. A., Shcherbatyy, A. R., Gutyj, B. V. Acid resistance and population structure of erythrocytes in trotter horses during and after exercise. *Regulatory Mechanisms in Biosystems*. 2017. Vol. 8. Iss. 4. P. 623–627. Doi: 10.15421/021795.

The share of "old" erythrocytes averaged 14.2±0.51% and was probably (p<0.05) higher than cows in the conditionally clean zone (12.5±0.33%). Since "old" forms of erythrocytes play an active role in oxygenation processes, the body will likely try to compensate for hypoxia by leaving these cells in the bloodstream.

The fraction of "mature" erythrocytes in cows from the contaminated zone averaged $37.7\pm0.76\%$ and was significantly (p<0.001) lower than in cows from the control group (43.4 \pm 0.40%). The "mature" erythrocyte population is functionally the most active; however, prolonged exposure to low doses of radiation leads to their aging.

The peak emergence on the experimental animals' erythrogram (Figure 1) starts from the 4th minute, 0.5 minutes later than the control. The peak height (hemolyzed cells) was 11.2% lower than in cows from the control group (17.0% compared to 28.2%).

The erythrocytes of experimental cows underwent maximum hemolysis at 5.5 minutes, while those of control cows experienced maximum hemolysis at 4.5 minutes. Destruction of erythrocytes was noted at the 9th and 7th minutes, respectively. Therefore, the acid resistance curve is characterized by a longer left part, which is an indicator of a more significant proportion of "old" erythrocytes in the blood of cows, and a stretched (longer) right part, which characterizes an increase in the proportion of "young" erythrocytes more resistant to hemolysis.

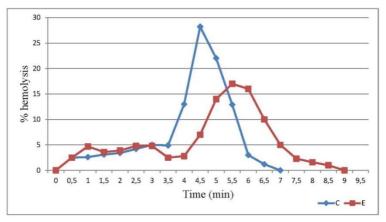


Fig. 1. Acid resistance of erythrocytes of cows in the zone of radioactive contamination

Therefore, the bodies of cows raised and kept on the farm, the land of which is contaminated with radionuclides, are constantly affected by factors that worsen the functional state of systems sensitive to the adverse effects of small doses of radiation, in particular, the hematopoietic system. Radiation pollution causes a violation of hematopoiesis and the development of anemia in cows, characterized by oligocythemia (90%) and oligochromemia (65%). The anemia is mainly hyperchromic (61.1%) or normochromic (38.9%) and macrocytic (66.7%). In cows, the erythrogram shows a longer left part, which indicates a significant number of "old" erythrocytes, and a longer right part, which characterizes an increased number of "young" erythrocytes.

Effective erythrocytopoiesis is possible, first of all, under the condition of optimal functioning of the red bone marrow, which is determined by the typical structural organization of its myeloid tissue, optimal provision of the body with nutrients (protein) and biologically active substances (cobalt, iron, copper, manganese, vitamins B_2 , B_6 , B_{12} , C).

A comparison of individual indicators of erythrocytopoiesis in cows of the experimental farm confirms the general pattern: hyperchromia in the vast majority of cows is combined with macrocytosis.

The effect of a lack of biotic microelements on hematopoiesis and other biological reactions is determined not only by their absolute amounts but also by their concentration in 1 kg of diet dry matter and an excess of antagonists.

With a lack of copper, hypochromic anemia should develop mainly, but in this economy – anemia is hyperchromic. It develops against the background of prevailing oligocythemia, the leading cause of which is a pronounced deficiency of cobalt in the diet, the supply of which is 49.9%. With a lack of cobalt, DNA synthesis in hematopoietic cells, particularly in erythro- and normoblasts, is reduced, and their division and maturation are delayed; macrocytic anemia develops ^{22, 23, 24, 25, 26, 27, 28}.

 22 Ветеринарна гематологія / О. І. Сукманський, С. І. Улизько ; за ред. проф. О. І. Сукманського. Одеса : ВМФ, 2009. 168 с.

²⁴ Shcherbatyy, A., Slivinska, L., & Lukashchuk, B. Hypocobaltosis and hypocuprosis in pregnant mares in the western biogeochemical zone of Ukraine (distribution, diagnosis). *Ukrainian Journal of Veterinary and Agricultural Sciences*. 2018. Vol. 1(2). C. 11–14.

²³ Shcherbatyy A. R., Slivinska L. G., Gutyj B. V., Fedorovych V. L., Lukashchuk, B. O. Influence of Marmix premix on the state of lipid peroxidation and indices of non-specific resistance of the organism of pregnant mares with microelementosis. *Regulatory Mechanisms in Biosystems*. 2019. Vol. 10(1). P. 87–91.

²⁵ Slivinska, L., Fedorovych, V., Gutyj, B., Lychuk, M., Shcherbatyy, A., Gudyma, T., Chernushkin, B., Fedorovych, N. The occurrence of osteodystrophy in cows with chronic micronutrients deficiency. *Ukrainian journal of Ecology*. 2018. Vol. 8. Iss. 2. P. 24–32. DOI: 10.15421/2018_305.

So, we found that the reason for the development of anemia in cows under the influence of radiation was not only the effect of low doses of ionizing radiation on the hematopoietic system but also the insufficient content of cobalt and copper in the diet and their low concentration in 1 kg of dry matter.

4. The antioxidant defense system (ADS) condition in cows under radiation exposure

The mechanism of radionuclides' action on living organisms is based on the phenomenon of disproportionation between the amount of absorbed energy of ionizing radiation and the magnitude of the biological effect it induces. This disproportionation leads to the formation of free radicals, which hurt living organisms^{29, 30, 31, 32}.

One of the critical elements in ensuring the functioning processes of a living organism belongs to the antioxidant defense system, which controls the level of free radicals formed through the participation of reactive oxygen species. Free radical processes play a significant role in the functioning of biological systems within normal limits. For instance, hydrogen peroxide participates in a complex series of reactions regulating cellular metabolism, and a slight increase in the concentration of $\rm O_2$ and $\rm H_2O_2$ can stimulate cell proliferation and other cellular functions. The activation of peroxidation processes plays a leading role in cell damage, and the accumulation of their products and highly toxic superoxide anions

Біологія тварин. 2006. Т. 8, № 1–2. С. 41–62.

²⁷ Судаков М., Береза В., Погурський І. Гіпокобальтоз: діагностика і профілактика в біогохімічних провінціях України Вет медицина України. 2000. № 8. С. 36–37.

²⁹ Слипанюк О. В. Антоняк Г. Л., Сологуб Л. І. Перекисне окиснення ліпідів і антиоксидантний статус у крові корів у останній місяць тільності. *Біологія тварин*. 2000. № 2(2). С. 83–86.

³¹ Зінко Г. О. Пероксидно-окисні процеси та стан системи антиоксидантного захисту у телят за гастроентериту. *Аграрний вісник Причорномор'я*. 2017. № 83. С. 86–90.

²⁶ Влізло В. В., Сологуб Л. І., Янович В. Г., Антоняк Г. Л., Янович Д. О. Біохімічні основи нормування мінерального живлення великої рогатої худоби. 2. Мікроелементи. *Біологія теалин* 2006 Т. 8. № 1–2. С. 41–62

В біогеохімічних провінціях України. Вет. медицина України. 2000. № 8. С. 36–37.

²⁸ Slivinska L. G., Lychuk M. G., Shcherbatyi A. R. Nutritional-deficiency anemia of cows. Prevalence and structure of metabolic diseases of laying hens. Topical issues of the development of veterinary medicine and breeding technologies. Scientific monograph. Riga, Latvia: "Baltija Publishing". 2022. P. 132–166.

³⁰ Shcherbatyy A. R., Slivinska L. G., Gutyj B. V., Fedorovych V. L., Lukashchuk, B. O. Influence of Marmix premix on the state of lipid peroxidation and indices of non-specific resistance of the organism of pregnant mares with microelementosis. *Regulatory Mechanisms in Biosystems*. 2019. Vol. 10(1). P. 87–91.

³² Gutyj, B., Stybel, V., Darmohray, L., Lavryshyn, Y., Turko, I., Hachak, Y., Shcherbatyy, A., Bushueva, I., Parchenko, V., Kaplaushenko, A., Krushelnytska, O. Prooxidant-antioxidant balance in the organism of bulls (young cattle) after using cadmium load. *Ukrainian journal of Ecology*, 2017. Vol. 7(4). P. 589–596.

leads to significant biochemical and biophysical disturbances in the body, exacerbating the negative course of pathological processes^{33, 34, 35}.

Prevention of the adverse effects of free radicals and peroxide compounds is ensured by a complex multistage antioxidant defense system (ADS). ADS is a complex of non-enzymatic antioxidants and specialized antioxidant enzymes^{36, 37}. Among the enzymatic antioxidants, particular importance is attributed to superoxide dismutase (SOD) and glutathione peroxidase (GPO). SOD is one of the first lines of defense mechanisms protecting the cell from the harmful effects of reactive oxygen species by neutralizing the superoxide radical, which initiates a series of free radical transformations. GPO reduces hydrogen peroxide hydroperoxides³⁸.

Considering the above, the study of radioecology in agricultural animals is necessary. It has been established that the interaction of O₂ with H₂O₂ is activated under radiation, leading to highly reactive hydroxyl radical OH, a potent activator of lipid peroxidation in cells^{39, 40, 41}.

Changes in animal lipid peroxidation processes are an essential mechanism under radiation exposure. Moreover, activation of the lipid oxidation system is one of the initiating mechanisms of the biological effects of ionizing radiation, playing a significant role in further manifestations

³³ Борисевич В. Борисевич Б., Борисевич В. (молодший). Вільні радикали і перекисне окиснення ліпідів у патогенезі хвороб тварин. Вет. медицина України. 2006. № 1. С. 15–17.

Антоняк Г. Л., Бабич Н. О., Сологуб Л. І., Снітинський В. В. Утворення активних форм кисню та система антиоксидантного захисту в організмі тварин. Біологія тварин. 2000. T.2, № 2. C. 34-43.

35 Слипанюк О. В. Антоняк Г. Л., Сологуб Л. І. Перекисне окиснення ліпідів і антиоксидантний статус у крові корів у останній місяць тільності. *Біологія тварин.* Львів, 2000. Т. 2, № 2. С. 83–86.

36 Слівінська Л. Г. Перекисне окиснення ліпідів у крові корів, хворих на хронічну

гематурію. *Наук. вісник Львів. нац. ун-ту вет. медицини та біотехнологій імені С. 3. І жиць-кого.* Львів, 2007. Т.9, № 3(34). Ч. 1. С. 154—159.

37 Левченко В. І., Слівінська Л. Г. Стан пероксидного окиснення ліпідів та системи

антиоксидантного захисту в корів, хворих на хронічну гематурію. Вісник Білоцерків. держ. аграр. ун-ту: зб.наук.праць. Біла Церква. 2007. Вип. 48. С. 93–97.

Слипанюк О. В., Антоняк Г. Л., Сологуб Л. І. Перекисне окиснення ліпідів і антиоксидантний статус у крові корів у останній місяць тільності. Біологія тварин. Львів, 2000. T. 2, № 2. C. 83–86.

9 Антоняк Г. Л., Бабич Н. О., Сологуб Л. І., Снітинський В. В. Утворення активних форм кисню та система антиоксидантного захисту в організмі тварин. Біологія тварин. 2000. T. 2. № 2. C. 34–43.

⁴⁰ Gutyj, B., Stybel, V., Darmohray, L., Lavryshyn, Y., Turko, I., Hachak, Y., Shcherbatyy, A., Bushueva, I., Parchenko, V., Kaplaushenko, A., Krushelnytska, O. Prooxidant-antioxidant balance in the organism of bulls (young cattle) after using cadmium load. Ukrainian journal of Ecology. 2017. Vol. 7(4). P. 589-596.

⁴¹ Shcherbatyy, A. R., Slivinska, L. G., Gutyj, B. V., Fedorovych, V. L., Lukashchuk, B. O. Influence of Marmix premix on the state of lipid peroxidation and indices of non-specific resistance of the organism of pregnant mares with microelementosis. Regulatory Mechanisms in Biosystems. 2019. Vol. 10. Iss. 1. P. 87-91.

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of radiation damage. Under the influence of radiation at low doses, disruptions in the antioxidant defense system (ADS) balance are possible, leading to uncontrolled lipid peroxidation (LPO) processes.

We have found that lipid hydroperoxides (LHP) content in the blood serum from farms in the conditionally clean zone averaged $2.08\pm0.084~UA~E_{480}/ml$. At the same time, in cows under radiation exposure, it was 24.0% higher (p<0.01) and ranged from 1.56 to 3.90 UA E_{480}/ml (Table 10).

Table 10

The condition of the lipid peroxidation-antioxidant defense system (LPO-ADS) in cows from the Rivne region under radiation exposure compared to those from the conditionally clean zone

		Gro	ups of animals	
Indicator	Biometric indicator	Conditionally clean zone, n=20	Radiation exposure, n=40	p<
LHP, UA E 480/ мл	Lim M±m	1,30–2,72 2,08±0,084	1,56–3,90 2,58±0,109	0,01
DC, μmol/L	Lim M±m	4,24–7,68 6,40±0,202	5,15-8,90 7,10±0,166	0,05
MDA, nmol/ml	Lim M±m	2,43-6,42 4,76±0,238	4,05–8,29 6,28±0,169	0,001
SOD, % protein. reaction/1 g of Hb	Lim M±m	0,215-0,446 0,334±0,016	0,178-0,382 0,262±0,010	0,001
GPO, µmol/min GSH per 1g of Hb	Lim M±m	208,5–395,6 304,1±15,97	152,2-258,6 199,0±4,10	0,001

Note: p< – compared to cows of a conditionally clean zone.

The content of DC in cows exposed to radiation was $7.10\pm0.166 \,\mu\text{mol/l}$ (lim 5.15-8.90), which is 10.9% (p<0.05) more, compared to cows in the conditionally clean zone, in which the average value of the indicator was $6.40\pm0.202 \,\mu\text{mol/l}$.

The concentration of MDA in the blood serum of cows in the conditionally clean zone was 4.76 ± 0.238 nmol/ml (2.43-6.42), and under the influence of radiation, it was 6.28 ± 0.169 nmol/ml, which is 31.9% more (p<0.001).

The concentration of free radicals in the cells of the body depends on the cooperative functioning of some enzymes, namely superoxide dismutase (SOD), which catalyzes the dismutation of superoxide anion radicals into hydrogen peroxide, and glutathione peroxidase (GPO), which catalyzes the reduction reaction of hydroperoxides using reduced glutathione.

We established low SOD activity in cows under radiation exposure of $0.262\pm0.010\%$ proteins. per 1 g of Hb (lim 0.178-0.382), which is 21.6% (p<0.001) less than in cows of the conditionally clean zone (0.334 $\pm0.016\%$ block reaction per 1 g of Hb). Such a reaction of SOD to the action of low doses of radiation may be associated with damage to the enzyme molecule by active products of PO.

The significant role in providing antioxidant protection belongs to GPO – a selenium-containing protein that catalyzes the reduction reaction of hydroperoxides using reduced glutathione. The average activity of GPO in cows from the conditionally clean zone was 304.1±15.97 μ mol/min GSH per 1g Hb, 52.8% higher (p<0.001) than in the radiation-affected zone. The enzyme activity in the latter ranged from 152.2 to 258.6, with an average value of 199.0±4.10 μ mol/min GSH per 1g Hb.

5. Correction of erythrocytopoiesis in cows with myelotoxic anemia in the zone of radiation contamination

Radiation stress has a significant negative impact on the health of animals in regions contaminated with radionuclides. The hematopoietic system is primarily affected by radiation exposure, and erythrocytopoiesis is significantly suppressed. The results of the research 42, 43, 44, 45 indicate that in the peripheral blood of cows whose bodies were exposed to radionuclides – strontium and cesium for ten years – oligocythemia, an increase in the color index of blood and the content of hemoglobin in erythrocytes was observed. Therefore, the development of measures to reduce the negative impact of radionuclides on erythrocytopoiesis is relevant for farms in the radiation pollution zone 46.

⁴² Демчук М. В., Висоцький А. О., Хміляр Д. Д. Морфологічні і окремі біохімічні показники крові корів при утриманні в зоні радіоекологічного контролю. *Екологія та проблеми зооінженерії і вет.медицини*. Харків, 1997. С. 37–38.

 43 Козенко О. В. Еритроцитарна система крові корів залежно від тривалості перебування в зоні, забрудненій радіонуклідами. *Наук. вісник Львів. держ. акад. вет.*

медицини імені С. З. Ґжицького. Львів, 2001. Т.З, № 4. Вип. 2. С. 27–30.

⁴⁶ Малина В. В., Козак М. В., Ткаченко Т. П., Нікітенко А. М. КАФІ – як фактор нормалізації гомеостазу телят при їх вирощуванні в зонах радіоактивного забруднення.

⁴⁴ Стронський Ю. С. Морфофункціональна характеристика червоного кісткового мозку молодняку великої рогатої худоби, вирощеного на забрудненій радіонуклідами території : автореф. дис. ... канд. вет. наук : спец. 16.00.02 "Патологія, онкологія і морфологія тварин". Київ, 2003. 18 с.

⁴⁵ Висоцький А. О. Вплив умов утримання на фізіологічний стан серцево-судинної системи і гемопоез у корів господарств зони радіоекологічного контролю : автореф. дис. ... канд. вет. наук : спец. 16.00.06 "Ветеринарна санітарія та гігієна". Львів, 2000. 20 с.

Our previous studies established that radiation pollution harmed hematopoiesis and caused the development of anemia in cows⁴⁷. In addition, studies have shown a deficiency in the diet of cobalt and copper (see Table 10). The results were a prerequisite for correcting erythrocytopoiesis and developing preventive measures for cow anemia.

Therefore, the next stage of the work was the study of indicators of erythrocytopoiesis in cows and the content of MD (Co, Fe, Cu) during the stable period of maintenance when feeding inorganic compounds of microelements and the complex drug developed by us, which includes inorganic and organic compounds (lactates) of MD. For research, 60 cows were selected on the farm, and three groups were formed: one control group and two experimental ones (n=20). The animals of the control group received the main diet (MD), and the first experimental group received inorganic compounds of microelements – sulfates of Cu, Zn, Mn, Fe, chloride of Co, KJ, and selenite of Na. The animals of the second experimental group were given a complex preparation containing organic compounds – lactates of Zn, Mn, Cu, Fe, Co, and Se salt on trilon and J starch. The experiment lasted 45 days.

While studying hematopoiesis indicators in the research groups' dry cows, it was established that inorganic compounds of microelements and their lactates positively affect hematopoietic processes (Figs. 1–4).

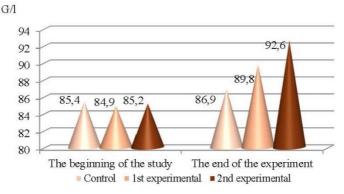


Fig. 2. Effect of ME salts and Mikrolact preparation on the hemoglobin content in the blood of cows

Наук. вісник Львів. держ. акад. вет. медицини імені С.З. Ґжицького. Львів, 2000. Т. 9, № 3(34). Ч. 1, С. 79–85.

⁴⁷ Слівінська Л. Г., Левченко В. І. Клінічний статус і показники гемопоезу лактуючих корів, вирощених на території, забрудненій радіонуклідами. *Сільський господар*. 2007. № 11–12. С. 33–36.

Hemoglobin content in the cows of the control group remained unchanged, and in the experimental ones, it tended to increase, especially in the cows of the second group. In the first group, the difference with the initial level was 5.8%, with the indicator in the control group -3.3%. In the second experimental group, the difference was 8.6 and 6.4%, respectively. At the end of the experiment, cows in the second group had 3.0% more hemoglobin than in the first, but the difference between the indicators was improbable (p<0.5).

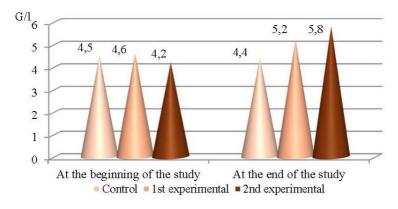


Fig. 3. Effect of ME salts and Mikrolact preparation on the number of erythrocytes in the blood of cows with myelotoxic anemia

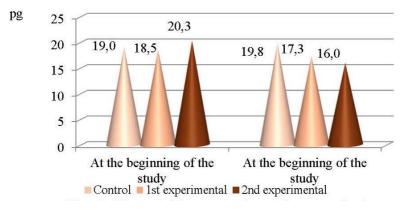


Fig. 4. Hemoglobin content in erythrocytes in the blood of cows

And yet, complete recovery of hemoglobin content did not occur, only in 7 (35 %) cows of the first experimental group and in 9 (45 %) of the second group, the hemoglobin content was within the limits of physiological fluctuations, and in the others it was less than the minimum standard limit. In 10 (50%) cows of the control group, this indicator was also below 95 g/l. Improbable changes in the hemoglobin content in the blood of the cows of the experimental groups are caused by wide limits both at the beginning and after the end of the experiment.

The number of erythrocytes before the start of MD feeding in the blood of cows of the first experimental group was 4.6 ± 0.19 T/l, the second -4.2 ± 0.20 T/l. In the control group, the number of erythrocytes was 4.5 ± 0.28 T/l, i.e., their number did not differ from the indicators in the cows of the experimental groups. After 45 days, no positive changes were found in this group's cows relative to the experiment's beginning.

After the end of the experiment (after 45 days), the number of erythrocytes increased significantly (p<0.05) in the blood of the cows of the first experimental group, compared to the beginning of the experiment and the control. The best results were obtained when feeding the complex drug in the cows of the second experimental group since the number of erythrocytes increased by 27.6 and 24.1% (p<0.001), compared to the beginning of the experiment and with the control, and in the cows of the first, by 13 and 24.1%, respectively 18.2%, that is, compared to the beginning, the difference between the groups was 14.6%.

Erythrocytes in the second group were significantly (p<0.05) more (by 11.5%) than in the first. If 40% of cows with oligocythemia remained in the first experimental group, then in the second -15 (in the control group -60%).

The increase in the number of erythrocytes and the tendency to increase the hemoglobin content in the cows of the experimental groups was influenced by the intake of cobalt and copper in the animals' bodies as part of the inorganic forms and lactate compounds of MD included in the main diet.

In the experimental cows, as a result of the treatment using MD salts and the drug, the number of erythrocytes increases more intensively than the hemoglobin content; therefore the saturation of erythrocytes with hemoglobin (MCH) decreases (Fig. 4): in the cows of the first experimental group by 14.4 and 7.5% according to the control and the beginning of the experiment (p<0.01–0.001), and after the use of MD lactates in the form of the drug, respectively, by 21.2 and 20.8% (p<0.001). It should be noted that in all cows of both experimental groups, the saturation of erythrocytes with hemoglobin was within the normal range. In contrast,

in the control group, 60% of cows had hyperchromic erythrocytes, which is, as a rule, the result of a lack of Co and vitamin B₁₂.

The amount of hematocrit (HNT) depends on the number of erythrocytes in the blood and their average volume. In 100% of cows of all three groups at the beginning of the experiment, it was reduced (Fig. 5), which is one of the indicators of anemia.

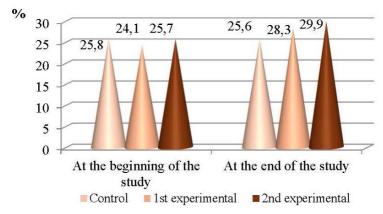


Fig. 5. The hematocrit value in cow blood

After the use of inorganic salts of MD and the drug Mikrolact in the blood of the cows of the experimental groups, this indicator probably (p<0.01) increased by 4.2 and 4.2%, respectively, relative to the beginning of the experiment, and by 2.7 and 4.3% (p<0.05 and 0.001) compared to the control. Nevertheless, we could not completely recover the hematocrit value in the cows of the experimental groups. More extended use of the drug is necessary since the recovery of the hematocrit value was in 60 and 85% of the cows of the experimental groups.

The hematocrit value is affected not only by the number of erythrocytes but also by their average volume, which was increased in some cows of the control and second experimental groups before the start of the experiment.

The use of micronutrient supplementation contributed to a decrease in the average volume of erythrocytes in cows of the second experimental group when fed Mikrolact, compared to the beginning of the experiment (p<0.001) and the control (p<0.01), which can be considered positive. In the blood of part of the cows of the control group, erythrocytes remained slightly more prominent in volume (macrocytes).

6. Correction of the exchange of trace elements in cows with myelotoxic anemia in the zone of radiation contamination

The soils and forages of the western regions of Ukraine contain insufficient biotic trace elements, which, together with other factors, cause anemia in cows. This problem was especially acute in farms in the zone exposed to radiation. As a result of the conducted research, it was established that animal feed use in regions of the country where the level of radionuclide contamination is high and the content of trace elements is low contributes to the accumulation of radionuclides in livestock products.

Our previous research established that the supply of beef cattle in the contaminated zone with trace elements in winter was insufficient. The most significant deficit in fodder was cobalt, zinc, and copper, and their supply to cows was 50.0%, 77.2, and 82.3% of the need, respectively. Only the level of manganese and ferrum in the diet of beef cows was high and amounted to 222.0 and 604.0% of the requirement, respectively.

Analyzing the copper content in cows' blood (Table 11), we found that it was lower than the minimum standard limit (14.1 μ mol/l) in 50 (83.3%) of the studied cows (hypocupremia), including 25 (50%) whose level was less than 13.0 μ mol/l. According to our research, the copper content in the blood of beef cows averaged 13.1 \pm 0.29 μ mol/l (9.8-16.3).

Table 11 The content of microelements in the blood of beef cows (n = 60)

Biometric indicators	Ferrum, µmol/l	Cobalt, µmol/l	Copper, µmol/l
Lim	17,5–29,4	0,241-0,421	9,8-16,3
M±m	$23,3\pm0,78$	$0,327\pm0,0110$	$13,1\pm0,29$
Standard	16,1–26,8	0,510-0,850	14,1–18,2

Copper deficiency in the animal body can manifest even at its optimal level in the diet due to factors that reduce its assimilation 48, 49, 50, 51.

⁴⁸ Sharma M. C., Joshi C., Pathak N. N., Kaur Copper H. Status and enzyme, hormone, vitamin and immune function in heifers. *Research in Veterinary Science*. 2005. Vol. 79, № 2. P. 113–123.

 $^{^{49}}$ Мінеральне живлення тварин / Г. Т. Кліценко, М. Ф. Кулик, М. В. Косенко [та ін.]. Київ, 2001. С. 5–44.

⁵⁰ Shcherbatyy, A., Slivinska, L., & Lukashchuk, B. Hypocobaltosis and hypocuprosis in pregnant mares in the western biogeochemical zone of Ukraine (distribution, diagnosis). *Ukrainian Journal of Veterinary and Agricultural Sciences*. 2018. Vol. 1(2). P. 11–14. https://doi.org/10.32718/ujvas1-2.03

⁵¹ Shcherbatyy, A. R., Slivinska, L. G., Gutyj, B. V., Golovakha, V. I., Piddubnyak, O. V., Fedorovuch, V. L. The influence of a mineral-vitamin premix on the metabolism of pregnant horses with microelemetosis. *Regulatory Mechanisms in Biosystems*. 2017. Vol. 8. Iss. 2. P. 293–298. Doi: 10.15421/021746.

A decrease in copper absorption occurs due to excess in the diet of ferrum and its antagonists, such as molybdenum, zinc, and sulfates. A decrease in its concentration in blood and tissues is observed in animals that received a ration with copper deficiency⁵².

Cuprum is necessary for converting ferrum into an organically bound form and plays an essential role in hemoglobin synthesis. By affecting the synthesis of iron-containing compounds, it can combine with some of them, forming iron-copper-nucleoprotein complexes, which are the precursors of hemoglobin. Cuprum contributes to the transfer of ferrum to the red bone marrow and is necessary for maturing reticulocytes into erythrocytes⁵³.

After the end of the experiment, the copper content in the blood of the cows of the 1st and 2nd experimental groups increased significantly (p<0.01; 0.001) by 14.2 and 20.8%, respectively, compared to the beginning of the experiment, and by 2.8 and 11.5%, compared to the control. In contrast, this indicator only tended to increase in the cows of the control group (Fig. 6).

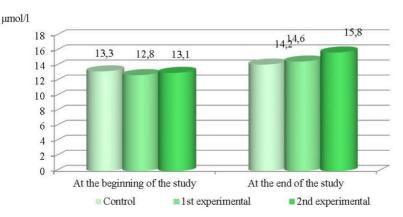


Fig. 6. The copper content in cow blood

⁵² Слівінська Л. Г., Щербатий А. Р., Личук М. Г. Вміст кобальту та купруму в молоці підсисних кобил гуцульської породи за мікроелементної корекції раціону. *Науковий вісник ЛНУВМ та БТ ім. С. 3. Гжицького.* Львів. 2014. Т. 16. № 3(60). Ч.1. С. 318–323.

⁵³ Щербатий А. Р. Діагностичні критерії та лікувально-профілактичні заходи за гіпокобальтозу і гіпокупрозу кобил гуцульської породи в біогеохімічній провінції Закарпаття [Текст] : автореф. дис... канд. вет. наук : 16.00.01. Білоцерк. нац. аграр. ун-т. Біла Церква, 2012. 20 с.

Cobalt is a synergist of copper and ferrum in the animal body. In their presence, its hematopoietic effect increases^{54, 55, 56}. Combined with copper and ferrum cobalt, it prevents oxidative and energy disorders in body tissues⁵⁷.

The cobalt content in the blood of the control group at the beginning of the experiment was on average $0.320\pm0.0106~\mu\text{mol/l}$, and in the cows of the first and second experimental groups -0.321 ± 0.0105 and $0.340\pm0.0096~\mu\text{mol/l}$, respectively (Fig. 7). In the blood of no cow, the cobalt content did not reach the lower limit of physiological fluctuations (0.51–0.85 μ mol/l), which is why they developed hyperchromic macrocytic anemia.

The content of Co in the first group after the end of the experiment increased to $0.49\pm0.018~\mu\text{mol/l}$ (+ 53.1%). Still, the average indicator did not reach the lower physiological limit (0.51 $\mu\text{mol/l}$) since only 30% of cobalt cows were within normal limits. The increase in Co content in the 2nd group was more significant: it reached $0.61\pm0.0245~\mu\text{mol/l}$ (+ 79.4% compared to the beginning of the experiment).

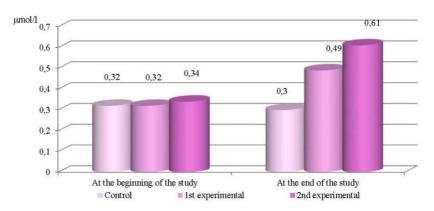


Fig. 7. Cobalt content in the blood of cows, μ mol/l

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⁵⁴ Судаков М., Береза В., Погурський І. Гіпокобальтоз: діагностика і профілактика в біогеохіміч-них провінціях України. *Вет. медицина України*. 2000. № 8. С. 36–37.

⁵⁵ Kincaid R. L., Lefebvre L. E., Cronrath J. D., Socha M. T., Johnson A. B. Effect of dietary cobalt supplementation on cobalt metabolism and performance of dairy cattle. *Journal of Dairy Science*, 2003. Vol. 86. P. 1405–1414.

⁵⁶ Личук М. Г. Роль нестачі селену та кобальту в кормах Полісся у виникненні мікроелементозів у телят: діагностика та лікування. *Наук. вісник Львів. держ. акад. вет. медицини імені С. З. Ґжицького.* 2000. Т. 3. № 2. Львів. С. 91–95.

⁵⁷ Слівінська Л. Г., Щербатий А. Р. Діагностика мікроелементозів кобил у західній біогеохімічній зоні України. *Вет. медицина України*. 2013. № 4(206). С. 25–28.

It was 26.3% more than in the 1st experiment group. According to the number of cows with a reduced Co level (25%), the effectiveness of Mikrolact is also more significant than that of inorganic salts of microelements (70%).

After the inclusion of MD inorganic compounds and the complex drug Mikrolact in the MD, the iron content in the blood of the experimental cows increased significantly (p<0.001) by 1.6 and 2.0 times, respectively, compared to the control and by 1.5 and 1.8 times (p<0.001) compared to the beginning of the experiment.

At the beginning of the experiment, the content of ferrum in the blood of animals of the control and experimental groups was within the physiological range (Fig. 8): in the cows of the control group, its content was, on average, $23.2\pm0.93~\mu\text{mol/l}$, in the 1st and 2nd and experimental groups -22.9 ± 0.72 and $23.8\pm0.70~\mu\text{mol/l}$, respectively. In 13 cows out of 60 examined (21.7%), its content was more significant than the upper limit of physiological fluctuations (26.8 μ mol/l). Due to an excessively high intake of ferrum in the body of animals, the assimilation of copper is inhibited⁵⁸.

According to the literature⁵⁹, ferrum absorption occurs mainly in the duodenum and depends on the saturation of ferritin of the intestinal mucosa and blood transferrin. The absorption process takes place in two stages: the capture of ferrum by the intestinal wall and its transport by the intestinal epitheliocyte into the blood. Organic acids that form insoluble iron salts (oxalates, citrates, possibly phytates) and an excess of zinc, manganese, copper and cadmium, phosphates, gossypol, and tannin inhibit iron absorption^{60, 61}.

After 45 days, the iron content in the blood serum of cows increased to $24.8\pm0.61~\mu\text{mol/l}$ in the first experimental group and to $26.1\pm0.43~\mu\text{mol/l}$ in the second, which was probably more by 7.7 (p<0.05) and 9.7% (p<0.05) compared to the beginning of the experiment, and by 6.0 and 11.5% (p<0.05) compared to with the control (23.4±0.91 μ mol/l).

⁵⁹ Антонюк Г. А., Сологуб Л. І., Снітинський В. В., Бабич Н. О. Залізо в організмі людини і тварин (біохімічні, імунологічні та екологічні аспекти). Львів, 2006. 310 с.

 $^{^{58}}$ Сологуб Л. І., Антоняк Г. Л., Стефанишин О. М. Роль міді в організмі тварин. Біологія тварин. 2004. Т.6, № 1–2. С. 64–76.

⁶⁰ Ostapyuk A. Yu., Gutyj B. V., Leskiv Kh. Ya., Shcherbatyi A. R. The toxic effect of cadmiumon the animal body and its prevention. *Achievements and research prospects in animal husbandry and veterinary medicine*: Scientific monograph. Riga, Latvia: «Baltija Publishing», 2023. 93–124.

⁶¹ Слівінська Л. Г., Щербатий А. Р., Личук М. Г., Гутий Б. В., Леньо М. І., Чернушкін Б. О., Приступа О. І., Драч М. П. (2023). Вплив кадмію і плюмбуму на стан антиоксидантної системи корів. *Матеріали науково-практичної онлайн конференції «Евзпечність та якість харчових продуктів у концепції «Єдине здоров'я»* (м. Львів, 1−2 червня 2023 р.). 102−103.

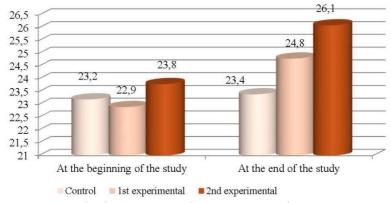


Fig. 8. Iron content in blood serum of cows

Thus, adding corrective additives of inorganic compounds of trace elements (1st group) and their lactates as part of the Mikrolact preparation (2nd group) positively affected the absorption and bioavailability of deficient trace elements, ensuring their optimal homeostasis. However, the best effect was obtained by including Mikrolact in the MD. In turn, inorganic MD salts and their lactates in the second experimental group contributed to restoring hematopoiesis indicators and eliminating hyperchromic macrocytic anemia. Lactates of microelements caused a more positive effect on all the investigated indicators than inorganic MD compounds.

7. The influence of inorganic compounds of trace elements on the course of metabolic processes in cows with myelotoxic anemia

Long-term keeping of animals in the territory contaminated with radionuclides causes changes in metabolic processes in the animal's body, in particular, in the protein composition of blood serum⁶². Therefore, one of the areas of study of the impact of trace elements on the animal body in the zone of radioactive contamination is the determination of protein fractions and enzyme activity in the blood.

The content of total protein in the blood serum of cows kept in the territory contaminated with radionuclides (control and two experimental groups) at the beginning of the study did not differ much: in cows of the control group it was 72.5 ± 1.38 g/l (62.4-85.2), the first test -74.1 ± 0.79 g/l

 $^{^{62}}$ Козенко О.В. Фізіологічний статус великої рогатої худоби за умов впливу абіотичних чинників середовища: автореф. дис...д-ра с.-г. наук: спец. 03.00.13 "Фізіологія людини і тварини" 16.00.06 "Гігієна тварин та ветеринарна санітарія". Львів, 2004. 41 с.

(69.3-81.4), the second -73.0 ± 1.31 g/l (62.5-83.8). Hypoproteinemia was established, respectively, in 25, 30 and 25% of the examined cows.

The albumin content in the blood serum of cows of the control, first, and second experimental groups was, on average, 27.5 ± 0.48 , 29.7 ± 0.53 , 29.1 ± 0.77 g/l, and their share in the total amount, respectively, 38.0 ± 0.29 ; 40.1 ± 0.70 and $39.8\pm0.41\%$.

A decrease in the share of albumins in 12 (60%) control cows and 9 (45%) cows in the experimental groups is an indicator of a violation of the albumin-synthesizing function of the liver or the filtering function of the kidneys.

Levels of α - and β -globulins in the blood serum of cows of all groups did not differ among themselves. The level of β -globulins in the blood serum was, on average, 9.3 ± 0.29 g/l in control, 10.0 ± 0.26 and 10.3 ± 0.29 g/l, respectively, in the first and second groups, and was within the physiological range. The content of γ -globulins in the blood serum of cows of the control and experimental groups was, on average, 27.2 ± 0.58 , 26.5 ± 0.62 and 25.6 ± 0.87 g/l, respectively.

The treatment carried out contributed to the normalization of the metabolic processes of the body of experimental cows due to the feeding of inorganic salts of microelements. Thus, the protein content in the blood serum of experimental cows after correction of the main diet with inorganic compounds of trace elements probably (p<0.01–0.001) increased by 5.8 and 8.2% compared to the beginning of the research and by 3.9 and 4.9% compared to control (p<0.05) in both the first and second groups.

The increase in the amount of total protein in the blood serum of cows occurred at the expense of the albumin fraction. In the cows of the first group, which were fed with inorganic salts of trace elements, the albumin content increased (p<0.01), compared to the beginning of the study by 13.5% and the control (+13.5%), of the second group, which were additionally fed to the main diet Mikrolact, respectively, by 19.9 and 17.5% (p<0.001). Since all albumins are synthesized exclusively by the liver, it can be asserted that feeding inorganic salts of microelements and the complex preparation Mikrolact has a positive effect on its albumin-synthesizing function. However, a somewhat better effect was obtained when feeding the complex preparation Mikrolact, compared to a group of cows given inorganic salts of trace elements. There was a tendency to increase the share of albumins (+1.1%; p<0.1) and their absolute amount. If, in the second group, it increased by 4% compared to the initial one and 4.6% compared to the control (p<0.001), less pronounced changes were observed in the first (+2.6 and 3.5%, respectively). Notably, the proportion of albumins in the blood serum of all cows of the second group was within the normal range (>38%).

Analyzing the fractional composition of globulins, we recorded the alpha fraction level in the control and experimental cows slightly increased. Still, there are no probable changes under the influence of MD compared to the control group. The content of beta-globulins in the blood serum of cows of the control and second experimental groups tended to increase. This increase was probable in the cows of the first group (p<0.05) compared to the beginning of the research. The increase was probable in both groups compared to the control group. The concentration of gamma globulins in all experimental groups tended to decrease, and their share in the total amount of protein probably decreased at the beginning of the study and concerning the indicator in the control group, in which similar changes were established (p<0.01).

In recent years, significant attention has been paid to the literature on determining the activity of liver indicator enzymes, in particular transferases.

The activity of AsAT in the blood serum of cows of the control and experimental groups was, on average, 2.39±0.10, 2.43±0.08 and 2.52±0.10 mmol/ (l×h), respectively. The enzyme activity was increased in 3 cows of the control group, two of the first, and 4 of the second experimental group. Since the activity of AsAT is relatively high in liver cells, even its slight increase in blood serum indicates damage to hepatocytes during an extended stay in the zone of radiation pollution ^{63, 64}.

After the end of the experiment, changes in the activity of AsAT in blood serum were characterized by a probable decrease in cows of the first (p<0.01) and second (p<0.001) groups. In contrast, only a trend (p<0.1) was noted in the cows of the control group since in the other two cows, its level exceeded the upper limit of physiological fluctuations (2.14 mmol/($l \times h$).

AlAT activity in the blood serum of cows at the beginning of the research ranged from 0.68–2.34 mmol/(l h) in the control group, 12.2–33.7 in the first group, and 0.85–1.93 mmol/ (l·h) in the second experimental groups. Enzyme activity was elevated in 3 cows of the control group and 4 of the first and second experimental groups.

After feeding the cows with inorganic compounds of trace elements and the Mikrolact preparation, the activity of AlAT in the cows of the first group tended to decrease compared to the beginning of the trial (p<0.1).

⁶⁴ Слівінська, Л. Г., Демидюк С. К., Щербатий А. Р. Синдроматика та стан метаболічних процесів у корів за мікроелементозів. *Науковий вісник Львівського національного університету ветеринарної медицини та біотехнологій імені С. З. Гжицького.* 2017.

T. 19. № 78. C. 182–186.

⁶³ Slivinska, L. G., Vlizlo, V. V., Shcherbatyy, A. R., Lukashchuk, B. O., Gutyj, B. V., Drach, M. P., Lychuk, M. G., Maksymovych, I. A., Leno, M. I., Rusyn, V. I., Chernushkin, B. O., Fedorovych, V. L., Zinko, H. O., Prystupa, O. I., Yaremchuk, V. Y. Influence of heavy metals on metabolic processes in cows. *Ukrainian Journal of Ecology*. 2021. Vol. 11(2). P. 284–291. doi: 10.15421/2021_112

In contrast, this difference was probable in the cows of the second experimental group (p<0.05). No significant difference was found in the blood serum of cows of the control group compared to the beginning of the experiment (p<0.5).

Thus, cows in the zone of radiation contamination develop anemic syndrome. The increased radioactive background increases the strength of the exposure dose of ionizing radiation, supplemented by the consumption of feed contaminated with radionuclides. The highest content of cesium-137 was in hay. Similar results are given by R.S. Dankovych⁶⁵, who also conducted research in the Dubrovytskyi district of the Rivne region. The negative impact of external and internal radiation on the body is supplemented by feeding animals with fodder with insufficient content of essential trace elements – cobalt, copper, iodine, and zinc. Cobalt and copper play a decisive role in the production and maturation of erythrocytes and the inclusion of ferrum in the heme molecule.

The listed factors are the reason for the development of hypoplastic anemia in 95.0% of cows, manifested by oligocythemia (95.0%) and oligochromemia (65%).

In addition to the decrease in the number of erythrocytes, we have established changes in their population composition: the number of "old" and "young" erythrocytes in the blood increases, and the proportion of "mature" decreases, which causes changes in their acid resistance.

The long-term effect of small radiation doses on the cows' bodies leads to significant deviations in the ADS. It harms the functions of the whole organism, which is constantly exposed to long-term ionizing influence.

In some cows, the albumin-synthesizing function of the liver is disturbed, although its changes are insignificant: the share of albumin in the total amount of protein is within 36.3–45.4%. Structural changes in hepatocytes are characterized by increased elimination into the bloodstream of liver indicator enzymes – aspartate and alanine aminotransferase.

To treat and prevent hypoplastic anemia in cows in the radioactive contamination zone, we tested two schemes for the use of essential trace elements (Co, Cu, Zn, Mn, Se, Fe): a) in the form of inorganic compounds of trace elements (the first research group); b) the use of the complex preparation Mikrolact, which includes Zn, Mn, Co, Cu, Fe lactates, and sodium selenite on trilon and starch iodine. The experiment showed the effectiveness of both MD application schemes, but we obtained better results in the second group (Table 12).

 $^{^{65}}$ Данкович Р. С. Морфологічна характеристика органів сечової системи великої рогатої худоби, вирощеної на забрудненій радіонуклідами території : автореф. дис. ... канд. вет. наук : спец. 16.00.02 "Патологія, онкологія і морфологія тварин". Київ, $2004.\ 20\ c.$

Table 12 Indicators of the effectiveness of the use of drugs containing MD

Indicator	The first group (n=20)	The second group (n=20)	p<	+ 2nd group to the first, in percentage
Hemoglobin, g/l	89,8±1,84	92,5±2,15	0,1	
Erythrocytes, T/l	5,2±0,15	5,8±0,18	0,05	11,4
Hematocrit, %	28,3±0,87	30,1±0,81	0,1	1,8
Co, μmol/l	$0,490\pm0,0182$	0,612±0,0245	0,01	24,9
Cu, μmol/l	14,62±0,38	15,83±0,46	0,05	8,3
Fe, µmol/l	24,8±0,61	26,1±0,43	0,1	5,2

Supplementation with microelement premixes increases the level of cobalt and copper in the blood of cows to a physiological level and eliminates the symptoms of anemia. The best therapeutic effect was observed in the animals of the second research group that received the drug Mikrolact, which maximally contributed to the increase in the level of cobalt and copper in the blood of cows, had a positive effect on the restoration of morphological indicators of blood, hemoglobin level, hematocrit value and saturation of erythrocytes with hemoglobin.

CONCLUSIONS

- 1. Conjunctival anemia was noted in 35.7% of cows from one of the farms in the Rivne region, which belongs to the third zone of radioactive contamination, 65% oligochromemia, and 90% oligocythemia. Anemia in 61.1% of cows is hyperchromic, and 66.7% is macrocytic. The population composition of erythrocytes is characterized by a decrease in "mature" and an increase in "young" and "old" fractions.
- 2. The drug Mikrolact in the zone contaminated with radionuclides contributed to an increase in the hemoglobin content by 8.6%, erythrocytes by 27.6%, stabilized the saturation of erythrocytes with hemoglobin and their volume, contributed to an increase in the content of copper and cobalt in the blood by 20.8 and 79.4% (15.8±0.96 and 0.61±0.0245 μ mol/l, respectively), of total protein (+19.9%) and the share of albumins (by 4%) and led to a decrease in the activity of AsAT and AlAT. Mikrolact was more effective in all parameters than inorganic MD salts.
- 3. Soils in the zone of technogenic load contain cadmium 3.6-4.8 times more than the maximum permissible concentration, lead -2.5-3.3, nickel 1.2-2.1, copper -5.9-7.0 times. The content of mobile forms of cadmium and lead increases at low pH of the soil environment. The most significant amount of cadmium and nickel accumulates in hay (coefficient 0.73-1.1),

- lead, and nickel in root crops. In the blood of cows in the zone of technogenic pollution, the cadmium content was $0.72\pm0.36-2.14\pm0.110$ µmol/l and was 4.3-12.6 times higher than that of cows in the control group (0.17 ± 0.010) , lead $-0.63\pm0.048-1.74\pm0.097$ µmol/l -1.43-3.96 times more than in control (0.44 ± 0.029) .
- 4. In 95% of cows in the zone of technogenic load with cadmium and lead compounds, normocytic anemia develops, mainly (73.7%) normochromic, less often hypo- or hyperchromic (23.7 and 2.6%). A negative correlation was established between the number of erythrocytes and the content of cadmium (r = -0.448) and lead (r = -0.434), the content of hemoglobin and cadmium (r = -0.64) and lead (r = -0.604). In the blood of cows, the proportion of "old" erythrocytes significantly increases (15.1±0.58 $-22.8\pm0.63\%$), and the population of "young" decreases (to 35.4±1.00%) compared to 14.2 ±0.39 and 43.2 $\pm0.54\%$, respectively, in cows of the control farm. Complete hemolysis of erythrocytes ends in them 1.0-2.0 minutes earlier.
- 5. The cobalt content is reduced in the blood of 100% of the cows in the technogenic load zone $(0.28\pm0.008-0.37\pm0.010~\mu\text{mol/l})$, 90% copper. A negative correlation was established between the content of cobalt and cadmium and lead (r=-0.545 and r=-0.550), copper and cadmium and lead (r=-0.671 and r=-0.646). Protein metabolism is characterized by the development of hypoproteinemia (in 32.5% of cows), hypoalbuminemia (30%), and hypogammaglobulinemia; 77.5% of cows have increased activity of AsAT, 66.5% AlAT. In the blood serum of cows in settlements located 3-5 km from the mine, the concentration of urea and creatinine increased.
- 6. In the body of anemic cows in the zone of artificial load, the processes of lipid peroxidation increase, and the content of their products in the blood increases, in particular MDA from 3.98±0.088 to 4.90±0.214 nmol/l, in the control 3.12± 0.122; the state of ADS is disturbed: the activity of SOD decreases (0.231±0.0051 % block.reaction/1 g Hb) and PLO (243.2±6.6 281.1±4.34 µmol/min GSH per 1 g Hb against 324.1±12.63 in control).
- 7. To treat cows and prevent myelotoxic anemia, inorganic compounds of microelements are used sulfates of copper, zinc, manganese, cobalt chloride, potassium iodide, and sodium selenite (the first group) and lactates of cobalt, copper, zinc, manganese, ferrum, sodium selenite on trilon and starch iodine (Mikrolakt preparation). The best therapeutic effect was established in the animals of the second group that received the drug Mikrolact, which contributed to the increase in the number of erythrocytes (28.7%), the content of hemoglobin (15.1%), copper (33.6%), and cobalt (2.3 times) had a positive effect on the restoration of the hematocrit value and saturation of erythrocytes with hemoglobin.

SUMMARY

Research conducted on dry cows, which were constantly kept in the zone of radioactive contamination, is presented. The soils of the diet are low in cobalt and copper (1.40 and 1.96 mg/kg). In the rations of the winter period, a deficiency of microelements, particularly copper and cobalt, was established: the supply is 82.4% and 49.9% of the need, and their concentration in 1 kg of dry matter was 5.7 and 0.24 mg. During the clinical examination of cows in the zone of radiation contamination, it was established that most of the animals had average fatness, dull hair cover, dry skin was observed in 63.8%, conjunctiva was pale pink in 35.7%, and mucous membranes of the nose and mouth were anemic. Tachycardia was recorded in 42.9% of cows and increased first heart sound – in 32.1%. Oligochromemia was recorded in 26 (65%) of the cows studied in the zone of radiation contamination, while in the cows of the conditionally clean zone, this indicator was within the physiological range. Oligocythemia was established in 90% of cows, and 65% was combined with oligochromemia. Cows in the zone of radioactive contamination mainly develop hyperchromic macrocytic anemia, the cause of which is not only the negative effect of low doses of ionizing radiation on the hematopoietic system but also, obviously, cobalt deficiency, which is characteristic of hypoplastic anemia. The share of "mature" erythrocytes is probably (p<0.001) smaller (by 5.7%) than in the control, and "old" erythrocytes, on the contrary, is higher by 1.7% (p<0.05). The curve of acid resistance of cows in the zone of radiation contamination is characterized by a longer left part, which is an indicator of a more significant number of "old" erythrocytes, and a longer right part, which is characteristic of an increase in the number of "young" erythrocytes: complete hemolysis ended in 9 minutes. Long-term exposure to small doses of radiation combined with a lack of biotic trace elements causes anemia in cows. It leads to significant deviations in the AOZ system, which has a negative effect on the body's function.

The inclusion of inorganic compounds of microelements and Mikrolact in the cows' diet caused an increase in the iron, copper, and cobalt in the blood of the cows in the experimental groups. In addition to the effect on hematopoiesis and the homeostasis of microelements, the applied drugs positively affect the course of metabolic processes, particularly the exchange of proteins. The total protein content in blood serum is restored due to an increase in the synthesis of albumins by 13.5 and 19.9% in the experimental groups (p<0.01 and p<0.001), and the activity of AsAT and AlAT decreases.

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